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(Sardinops sagax)
OFF CALIFORNIA IN 2005

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U.S. DEPARTMENT OF COMMERCE
National Oceanic and Atmospheric Administration
National Marine Fisheries Service
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SUMMARY

The spawning biomass in 2005 of Pacific sardine (*Sardinops sagax*) was estimated to be 621,657 metric tons (mt, CV = 0.54) for an area of 253,620 km^2 off California from San Diego to San Francisco, using the daily egg production estimate of 1.916/0.05m^2 (CV = 0.416) and the daily specific fecundity of 15.67 (number of eggs/population weight (g)/day). The area (253,620 km^2) was smaller than those in previous years: 320,620 km^2 (2004) and 365,906 km^2 (2003). In 2005, trawl samples were taken in both high (Region 1) and low (Region 2) sardine egg density areas to ensure the accuracy of the adult reproductive parameters estimates. Examinations of results indicate that spawning biomass may have been underestimated in past years when trawl samples were only taken from Region 1. In addition, when the 14 trawls taken in 2005 were classified as inshore or offshore, female sardines were found to be larger and to spawn more frequently offshore than inshore. The estimates of spawning biomass of Pacific sardine in 1994 - 2005 are 127,000 mt, 80,000 mt, 83,000 mt, 410,000 mt, 314,000 mt, 282,000 mt, 1.06 million mt, 791,000 mt, 206,000 mt, and 485,000 mt, 300,000 mt, and 600,000 mt, respectively. Therefore, the estimates of spawning biomass have been fluctuating and have been higher in most years since 1994. The time series of spawning biomass starting from 1985 is one of the fishery-independent inputs to the annual stock assessment of the Pacific sardine.

INTRODUCTION

The spawning biomass of Pacific sardine (*Sardinops sagax*) during 1986 (Scannel et al. 1996), 1987 (Wolf 1988a), 1988 (Wolf 1988b), 1994 (Lo et al. 1996), and 1996 (Barnes et al. 1997) was estimated independently using the daily egg production method (DEPM: Lasker 1985). The DEPM estimates spawning biomass by: 1) calculating the daily egg production from ichthyoplankton survey data, 2) estimating the maturity and fecundity of females from adult fish samples, and 3) calculating the biomass of spawning adults. Before 1996, sardine egg production was estimated from CalVET plankton net samples. Adult fish were sampled in various ways prior to 1996 to obtain specimens for batch fecundity, spawning fraction, sex ratio, and average female fish weight (Wolf 1988a, 1988b; Scannell et al. 1996; Macewicz et al. 1996; Lo et al. 1996).

Since 1996, in addition to CalVET and Bongo nets, the Continuous Underway Fish Egg Sampler (CUFES; Checkley, et al. 1997) has been used as a routine sampler for fish eggs, and data of sardine eggs collected with CUFES have been incorporated in various ways depending on the survey design in the estimation procedures of the daily egg production. In the 1997 sardine egg survey (Hill et al. 1998, Lo et al. 2001), CUFES was used to allocate CalVET tows in an adaptive sampling plan. From 1998 to 2000, data of sardine eggs collected with both CalVET and CUFES during each April CalCOFI cruise were used to estimate daily egg production (Hill et al. 1999). Use of the full data sets from both samplers in the DEPM can be time consuming. Furthermore, the CUFES samples are exclusively from 3 m depth and it is not clear whether the distributions of sardine egg stages from CUFES samples are representative. Use of the CUFES data also requires an estimated conversion factor from eggs/min to eggs/0.05m^2. Starting with the 1999 April CalCOFI survey, an adaptive allocation survey design similar to the 1997 survey was implemented. In this design, CalVET tows are added in areas where they were not preassigned if sardine egg densities in CUFES collections are high.

Since 2001, a cost-effective alternative has been adopted to retain the DEPM index, but in a revised form that reduces effort in calculation and egg staging for CUFES collections. This revised DEPM index only uses CalVET samples of eggs and yolk-sac larvae and Bongo samples of yolk-sac larvae in the high density area (Region 1) to provide an estimate of P_0, the variance of which can be large due to small sample size (fewer than 100 plankton tows).

Since 2004, a full-scale survey has been conducted for collection of sardine eggs, larvae, and adults to estimate the spawning biomass of Pacific sardine (Lo and Macewicz 2004). Unlike the survey in 2004 when the adult samples were taken primarily in the high density area, in 2005 adult sardine samples for reproductive output were taken in both high and low density areas aboard the NOAA ship *David Starr Jordan*. The ichthyoplankton samples were taken aboard the R/V *New Horizon* and the R/V *David Starr Jordan*.

MATERIALS AND METHODS

Data

Sardine eggs collected with both CalVET and CUFES during the April 2005 *New Horizon* and *Jordan* cruises were the data sources for estimating the daily egg production of sardine. In addition to sardine eggs and yolk-sac larvae collected with the CalVET net, yolk-sac larvae collected with the Bongo net have been included to model the sardine embryonic mortality curve since 2000. Since 2001 (Lo 2001), the CUFES data from the ichthyoplankton surveys have been used only to map the spatial distribution of the sardine spawning population with the survey area post-stratified into high density and low density areas according to the egg density from CUFES collections. Staged eggs from CalVET tows and yolk-sac larvae from CalVET and Bongo tows in the high density area have been used to model embryonic mortality curve in the high density area and later converted to the daily egg production, P_0, for the whole survey area.

During the 2005 survey, the regular CalCOFI survey was extended to CaCOFI line 60.0 (San Francisco). The *New Horizon* cruise (April 15 - May 1) occupied the six regular CalCOFI lines (93.3 - 76.6) and the *Jordan* cruise (March 28 - April 26) occupied 13 additional lines (94.7- 60.0). Therefore, a total of 19 lines was occupied by both vessels, with lines being 20 or 40 nm apart (Figure 1). Bongo samples were taken only at regular CalCOFI survey stations aboard both the R/V *New Horizon* in the south and the R/V *David Starr Jordan* in the north. On the southern 6 lines CalVET tows were taken only at regular CalCOFI survey stations. For the 13 *Jordan* lines, CalVET tows were taken at 4 nm intervals on each line after the egg density from each of two consecutive CUFES samples exceeded 1 egg/min and CalVET tows were stopped after the egg density from each of two consecutive CUFES samples was less than 1 egg/min. The threshold of 1 egg/min was reduced from the number used in years prior to 2002 (2 eggs/min) to increase the area identified as the high density area and, subsequently, to increase the number of CalVET samples. This adaptive allocation sampling was similar to the 1997 survey (Lo et al. 2001).

The survey area was post-stratified into two regions: Region 1, the high density area, and Region 2, the low density area. Region 1 encompassed the area where the egg density in CUFES collections was at least 1 egg per minute. The sizes of Region 1 and the total survey area were calculated using the formula for trapezoid area. The area of Region 1 was 46,203 km^2, which is 18% of the total survey area of 253,620 km^2. The rest of the survey area was Region 2 (Figure 1). One egg/min is equivalent to two to seven eggs/CalVET tow, depending on the degree of water mixing.

A total of 961 CUFES samples was collected from both the *New Horizon* (399) and *Jordan* (562) cruises. CUFES sampling intervals ranged from 1 to 60 minutes with a mean of 26 minutes and median of 30 minutes. A total of 74 CalVET samples was collected, of which 23 contained at least one sardine egg (Table 1). Egg densities from each CalVET sample and from the CUFES samples taken within an hour before and after the CalVET tow, were paired and used to derive a conversion factor (*E*) from eggs/min of CUFES sample to CalVET catch. We used a regression estimator to compute the ratio of mean eggs/min from CUFES to mean eggs/tow from CalVET: $E = \mu_y / \mu_x$ where y is the eggs/min and x is eggs/tow.

Daily egg production (P_0)

Similar to the 2001-2004 procedure (Lo 2001), we used the net tow as the sampling unit. Eggs from CalVET tows and yolk-sac larvae from both CalVET and Bongo tows in Region 1 were used to compute egg production primarily based on data from 11 transects (lines 93-75) (Figure 1). A total of 18 of the 26 CalVET samples in this region contained ∃1 sardine egg; these eggs were examined for their developmental stages (Figure 2).

Based on aboard-ship counts of eggs in CUFES samples, 297 of the 961 collections were positive for sardine eggs. In Region 1, there were 155 positive CUFES collections out of 188 total collections. In Region 2, 142 of the total 773 collections were positive (Table 1).

For modeling the embryonic mortality curve, yolk-sac larvae (larvae #5 mm in preserved length) were included assuming the mortality rate of yolk-sac larvae was the same as that of eggs (Lo 1986). Yolk-sac larval production was computed as the number of yolk-sac larvae/0.05m^2 divided by the duration of the yolk-sac stage (number of larvae/0.05m^2/day), and the duration was computed based on the temperature-dependent growth curve (Table 3 of Zweifel and Lasker 1976) for each tow. For yolk-sac larvae caught by the Bongo net, the larval abundance was further adjusted for size-specific extrusion from 0.505 mm mesh (Table 7 of Lo 1983) and for the percent of each sample that was sorted. The adjusted yolk-sac larvae/0.05 m^2 was then computed for each tow and was termed daily larval production/0.05 m^2. In the entire survey area, 16 of 74 CalVET and 33 of 101 Bongo samples had at least one yolk-sac larva (Figure 3). In Region 1, 10 of 23 CalVET and 11 of 15 Bongo samples were positive for yolk-sac larvae. In Region 2, 6 of 51 CalVET and 22 of 86 Bongo samples were positive for yolk-sac larvae (Table 1).

Daily egg production in Region 1 ($P_{0,1}$)

Sardine eggs and yolk-sac larvae and their ages were used to construct an embryonic mortality curve (Lo et al. 1996). Sardine egg density for each developmental stage was computed based on CalVET samples (Figure 2). The density of eggs in 2005 was similar to that in 2004, lower than that in 2003 and similar to 2002 (Lo and Macewicz 2002, Lo 2003). Unlike most of the past data where the density of eggs in stages 6 was highest among all stages, the density of eggs in stage 3 was highest in 2005. A temperature-dependent stage-to-age model (Lo et. al. 1996) was used to assign age to each stage. Sardine eggs and estimated ages were used directly in nonlinear regression. Eggs ≤ 3-h old and eggs older than 2.5 days were excluded because of possible bias. The average sea surface temperature for CalVET tows with ∃1 egg was 14.2˚C, higher than in 2004 (13.4˚C) or in 2003 (13.8˚C), and close to the 14.1˚C average in 2000.

The sardine embryonic mortality curve was modeled by an exponential decay curve (Lo et al. 1996):

$$P_t = P_0 e^{-zt} \qquad [1]$$

where P_t is either eggs/0.05m^2/day from CalVET tows or yolk-sac-larvae/0.05m^2/day from

CalVET and Bongo tows, and *t* is the age (days) of eggs or yolk-sac larvae from each tow. A weighted nonlinear regression was used to estimate two parameters in equation (1) where the weights were 1/SD. The standard deviation (SD) of eggs was 30.5, 5.3, and 3.67, for day one, day two and day three age groups respectively. The high SD for the day one age group was due to a high catch of 178 eggs, with 155 eggs in stage 3 (10 hr old), at a sea surface temperature of 14.3˚C. The SD of yolk-sac larval production from CalVETs was 0.66 and the SD of yolk-sac larval production from Bongo samples was 2.5.

A simulation study (Lo 2001) indicated that $P_{0,1}$ computed from a weighted nonlinear regression based on the original data points has a relative bias (RB) of -0.04 of the estimate where the RB = (mean of 1,000 estimates - true value)/mean of 1,000 estimates. Therefore the bias-corrected estimate of egg production in Region 1: $P_{0,1,c} = P_{0,1} * (1- \text{RB}) = P_{0,1} *(1.04)$, and $\text{SE}(P_{0,1,c}) = \text{SE}(P_{0,1}) * 1.04$.

Daily egg production in Region 2 ($P_{0,2}$)

Although 48 CalVET samples were taken in Region 2, only 5 tows had ∃1 sardine egg, ranging from 1 to 7 eggs per tow (Table 1). Therefore, we estimated daily egg production in Region 2 ($P_{0,2}$) as the product of the bias-corrected egg production in Region 1 ($P_{0,1,c}$) and the ratio of egg density in Region 2 to Region 1 (***q***) from CUFES samples, assuming the catch ratio of eggs/min from CUFES to eggs/tow from CalVET is the same for the whole survey area:

$$P_{\mathbf{0,2}} = P_{\mathbf{0,1,}c}q \qquad [2]$$

$$q = \frac{\sum_i \frac{\bar{x}_{2,i}}{\bar{x}_{1,i}} m_i}{\sum_i m_i} \qquad [3]$$

$$\text{var}(q) = \frac{[n/(n-1)]\sum_i m_i^2 (q_i - q)^2}{\left(\sum_i m_i\right)^2}$$

where ***q*** is the ratio of eggs/min between the low density and high density areas, m_i was the total CUFES time (minutes) in the ith transect, $\bar{x}_{j,i}$ is eggs/min of the ith transect in the jth Region, and $q_i = \frac{\bar{x}_{2,i}}{\bar{x}_{1,i}}$ is the catch ratio in the ith transect.

Daily egg production for the whole survey area (P_0)

P_0 was computed as the weighted average of $P_{0,1}$ and $P_{0,2}$:

$$P_0 = \frac{P_{0,1,c}A_1 + P_{0,2}A_2}{A_1 + A_2} = P_{0,1,c}w_1 + P_{0,2}w_2 = P_{0,1,c}[w_1 + qw_2] \quad [4]$$

and

$$mse(P_0) = mse(P_{0,1,c})(w_1 + w_2 q)^2 + P_{0,1,c}^2 w_2^2 V(q) - mse(P_{0,1,c})w_2^2 V(q)$$

(Goodman 1960) where $mse\ (P_{0,1,c}) = v(P_{0,1}) + bias^2 = v(P_{0,1}) + (P_{0,1}\ RB)^2$

and $w_i = \frac{A_i}{A_1 + A_2}$, and A_i is the area size for i = 1 or 2.

Adult parameters

The trawl survey was conducted from March 28 to April 26 aboard the NOAA ship *David Starr Jordan* during the April CalCOFI ichthyoplankton survey (Figure 4). We conducted trawling near the surface (0-6 fathoms depth) at night in potential adult sardine areas as identified by the presence of sardine eggs in CUFES collections. A total of 19 trawls was taken of which 14 were positive for sardines. Adult Pacific sardines were dispersed or in small schools.

Up to 50 sardines were randomly sampled from each positive trawl (Table 2). If necessary, additional mature females were collected to obtain 25 mature females per trawl for reproductive parameters or for use in estimating batch fecundity. Each fish was sexed, standard length (mm) and weight (g) were measured, otoliths were removed for aging, and ovaries were removed and preserved in 10% neutral buffered formalin. Each preserved ovary was blotted and weighed to the nearest milligram in the laboratory. Ovary wet weight was calculated as preserved ovary weight times 0.78 (unpublished data, CDFG 1986). A piece of each ovary was removed and prepared as hematoxylin and eosin (H&E) histological slides. All slides were analyzed for oocyte development, atresia, and postovulatory follicle age to assign female maturity and reproductive state (Macewicz et al. 1996).

Four adult parameters are needed for estimation of spawning biomass: 1) daily spawning fraction or the number of spawning females per mature female per day (S); 2) the average batch fecundity (F); 3) the proportion of mature female fish by weight (sex ratio, R); and 4) the average weight of mature females (g, W_f). Population values for S, R, F and W_f were estimated by methods in Picquelle and Stauffer (1985). Daily specific fecundity (number of eggs per population weight (g) per day) is RSF/W_f. Correlations among all pairs of adult parameters were calculated for computing the variance of the estimate of spawning biomass (Parker 1985). An

MS ACCESS[1] Visual Basic program (Chen et al. 2003) was used to summarize the trawl adult parameters, calculate adult parameter correlations, and to estimate spawning biomass and covariance.

Spawning fraction (S). A total of 175 mature female sardines was analyzed and considered to be a random sample of the population in the area trawled. Histological criteria can be used to identify four different spawning nights: postovulatory follicles aged 44-54 hours old indicated spawning two nights before capture (A); postovulatory follicles aged about 20-30 hours old indicated spawning the night before capture (B); hydrated oocytes or new (without deterioration) postovulatory follicles indicated spawning the night of capture (C); and early stages of migratory-nucleus oocytes indicated that spawning would have occurred the night after capture (D). The daily spawning fraction can be estimated by using the number of females spawning on one night, an average of several nights, or all nights. We used the number of females identified as having spawned the night before capture (B) and the adjusted number of mature females caught in each trawl (Table 2) to estimate the population spawning fraction and variance which is the default spawning night in the EPM program (Chen et al. 2003) and the traditional method of Picquelle and Stauffer (1985).

Batch fecundity (F). Batch fecundity (number of oocytes per spawn) was considered to be the number of migratory-nucleus-stage oocytes or the number of hydrated oocytes in the ovary (Hunter et al., 1985). We used the gravimetric method (Macewicz et al. 1996; Hunter et al. 1985, 1992) to estimate mean batch fecundity for 22 females caught during the April 2005 survey. The relationship of batch fecundity (F_b) to female weight (without ovary, W_{of}), as determined by simple linear regression, was F_b = -6085 + 376.28W_{of} where r^2 = 0.948 and W_{of} ranged from 33 to 193g (Figure 5). We used this equation to predict batch fecundity for each of the 175 mature Pacific sardine females analyzed to estimate spawning frequency.

Female weight (W_f). The observed female weight was adjusted downward for females with hydrated ovaries because their ovary weights were temporarily inflated. We obtained the adjusted female weight by the linear equation W_f = -1.88 + 1.06W_{of} where W_f is wet weight and W_{of} is ovary-free wet weight based on data from non-hydrated females taken during the April 2005 survey.

Sex ratio (R). The female proportion by weight was determined for each trawl (or each collection). The average weight of males and females (calculated from the first 10 males and 25 females) was multiplied by the number of males or females in the collection of 50 randomly selected fish to calculate total weight by sex in each collection. Thus, the female proportion by weight in each collection (Table 2) was calculated as estimated total female weight divided by estimated total weight in the sample. The estimate of the population's sex ratio by weight was calculated (Picquelle and Stauffer, 1985).

[1] Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

Spawning biomass (B_s)

The spawning biomass was computed according to:

$$B_s = \frac{P_0 AC}{RSF / W_f} \qquad [5]$$

where A is the survey area in unit of 0.05 m^2, S is the number of females spawning per mature females per day, F is the batch fecundity (number of eggs per mature female), R is the fraction of mature female fish by weight (sex ratio), W_f is the average weight of mature females (g), and C is the conversion factor from grams (g) to metric tons (mt). P_0A is the total daily egg production in the survey area, and the denominator (RSF/W_f) is the daily specific fecundity (number of eggs/population weight (g)/day).

The variance of the spawning biomass estimate $(\hat{B}_s)$ was computed from the Taylor expansion and in terms of the coefficient of variation (CV) for each parameter estimate and covariance for adult parameter estimates (Parker 1985):

$$VAR(\hat{B}_s) = \hat{B}_s^{\,2}\left[CV(\hat{P}_0)^2 + CV(\hat{W}_f)^2 + CV(\hat{S})^2 + CV(\hat{R})^2 + CV(\hat{F})^2 + 2COVS\right] \qquad [6]$$

The last term involving covariance term on the right-hand side is

$$COVS = \sum_i \sum_{i<j} sign \frac{COV(x_i, x_j)}{x_i x_j}$$

where x's are the adult parameter estimates, and subscripts i and j represent different adult parameters; e.g., $x_i = F$ and $x_j = W_f$. The sign of any two terms is positive if they are both in the numerator of B_S or denominator of B_S (equation 5); otherwise, the sign is negative. The covariance term is

$$\mathrm{cov}(x_{i,} x_j) = \frac{[n/(n-1)]\sum_k m_k (x_{i,k} - x_i) g_k (x_{j,k} - x_j)}{\left(\sum_k m_k\right)\left(\sum_k g_k\right)}$$

where k refers to k^{th} tow, and k=1,…,n. The terms of m_k and g_k are sample sizes and $x_{i,k}$ and $x_{j,k}$ are sample means from the k^{th} tow for x_i and x_j respectively.

RESULTS

Daily egg production (P_0)

In Region 1, the daily egg production ($P_{0,1}$) (equation 1) was 7.84/0.05 m^2/day (CV = 0.40) compared to 3.78/0.05 m^2/day in 2004 (CV = 0.23), egg mortality was Z = 0.58 (CV = 0.2) compared to 0.25 (CV=0.04) in 2004, and the area was 46,203 km^2 (13,500nm^2) compared to 68,203 km^2 (19,928 nm^2) in 2004 (equation 1 and Figure 6). The bias-corrected egg production, ($P_{0,1,c}$) is 8.14 (CV = 0.4) (Table 3). The ratio (***q***) of egg density between Region 2 and Region 1 from CUFES samples was 0.065 (CV=0.61) (equation 3). In Region 2, the egg production ($P_{0,2}$) was 0.53 /0.05 m^2/day (CV = 0.69) for an area of 207,417 km^2 (60,604 nm^2). The estimate of the daily egg production for the entire survey area was 1.916/0.05 m^2 (CV = 0.42) (equation 4) for a total area of 253,620 km^2 (74,104 nm^2) (Table 3). Egg mortality was similar to rates in years 2000-2003 (Table 4).

Catch ratio between CUFES and CalVET (E)

Although this ratio is no longer needed in the current estimation procedure, we computed it for comparison purposes. The catch ratio of eggs/min to eggs/tow (eggs/min = E * eggs/0.05 m^2) was computed from 22 pairs of CalVET tows and CUFES collections, excluding the maximum CalVET tow (178 eggs) as the maximum of eggs in all other tows was fewer than 50 (Figure 7). The eggs/min corresponding to each positive CalVET tow was the mean of all CUFES collections taken from one hour before to one hour after each positive CalVET tow. The catch ratio was 0.18 (CV = 0.28) compared to the 2004 estimate of 0.22 (CV = 0.09) and the 2003 estimate of 0.39 (CV = 0.11). A ratio of 0.18 means that one egg/tow from a CalVET tow was equivalent to approximately 0.18 egg/min from a CUFES sample, or one egg/minute from the CUFES was equivalent to 5.55 eggs/tow from the CalVET sample.

Adult parameters

Standard length (SL) of the first 50 randomly selected sardine in each trawl ranged from 118 to 258 mm for 277 males and from 121 to 265 mm for 229 females; one 249 mm sardine was not sexed because the gonad no longer existed. 32% of the females were immature and ranged from 121 to 182 mm SL. Using logistic regression (Macewicz et. al 1996, Lo et al. 2005), the length at which 50% of females are mature (ML_{50}) was calculated as 152.1 mm SL (Figure 8).

Reproductive parameters of 175 mature female sardines (up to 25 mature analyzed per trawl) for the individual trawls are given in Table 2. The April 2005 population sex ratio (R), was 0.469 (CV = 0.07) (Table 5). Estimates of the other female sardine parameters were: F, mean batch fecundity, was 17,662 eggs/batch (CV = 0.17); S, spawning fraction, was 0.124 per day (CV = 0.31); and W_f, mean female fish weight, was 65.34 grams (CV = 0.11). The average interval between spawning (spawning frequency) was about 8 days (inverse of spawning fraction or 1/0.124), and the daily specific fecundity was 15.67 eggs/gm/day (Table 5). The correlation matrix for the adult parameter estimates is shown in Table 5.

Spawning biomass (B_s)

The final estimate of spawning biomass of sardine in 2005 (equation 5, Table 5) was 621,657 mt (CV=0.54) or 683,823 short tons (st) (=621,657 x 1.1) for an area of 253,620 km^2 (74,104 nm^2) from San Diego to San Francisco. The point estimates of spawning biomass of Pacific sardine in 1994-2004 are respectively 127,102; 79,997; 83,176; 409,579; 313,986; 282,248; 1,063,837; 790,925; 206,333; 485,121 and 281,639 mt (Table 4).

DISCUSSION

Sardine eggs

Sardine eggs were concentrated between CalCOFI lines 87 and 75. The distribution of sardine eggs was different from the past two years because very few were collected north between lines 73 and 60 (Figure 1). The egg density (0.62 egg/min, Table 3) was similar to that in 2004 (0.78 egg/min, Lo and Macewicz 2004) and lower than that in 2003 (1.57 egg/min, Lo 2003). There seems to be less spawning activity in the southern part of the survey area since 2002, especially off San Diego.

The adaptive allocation sampling procedure was used only aboard the NOAA ship *David Starr Jordan*, which covered the area between CalCOFI lines 95 and 60, and not aboard the R/V *New Horizon*, because the latter was conducting the routine CalCOFI survey. Even though a high percent of the positive CalVET tows taken during the *New Horizon* CalCOFI cruise were in the high density area (10 positive tows out of 15), additional CalVET tows that the adaptive sampling procedure would have allocated were not taken. As a result, the 74 total CalVET tows was lower than that taken in 2004 (n = 124). Again, we highly recommend that the adaptive allocation sampling be applied aboard the research vessel that conducts the spring (March-April) routine CalCOFI survey in the future to ensure the quality of the estimate of the spawning biomass of Pacific sardine.

Embryonic mortality curve

The estimates of the daily egg production at age 0 (P_0/0.05 m^2) and the daily embryonic mortality were higher than in previous years. These high values were not caused by high abundance of eggs, but by the distribution of egg developmental stages (Figure 2). In 2005, the peak density among egg developmental stages was that of stage 3, rather than the older stages 5 or 6, seen in past years. The latter phenomenon is not understood and needs thorough investigation. Another observation is that the coefficients of variation (CV) of both P_0 and Z were higher than any of those obtained in the past. This high CV may be the result of smaller sample size of CalVET tows (74) in 2005 than in previous years: 217 in 2002, 192 in 2003 and 124 in 2004. The values of high CVs were similar to those in 2000 when the estimates and CVs of P_0 and Z were highest among all years.

Catch ratio between CUFES and CalVET (E)

The 2005 catch ratio between CUFES and CalVET (0.18) was lower than most of those obtained in recent years: 1998 (0.32), 1999 (0.34), 2000 (0.277), 2001 (0.145(CV = 0.026)), 2002 (0.24(CV = 0.06)), 2003 (0.39(CV = 0.11)), and 2004 (0.22 (CV = 0.09)). This 2005 value was quite different from the 1996 estimate of 0.73. This could be because the 1996 CalVET samples were taken only in the southern area near San Diego while after 1997 CalVET samples were taken in a larger area extending far north of San Diego.

Adult parameters

During the 2005 sardine survey, we were able to conduct some trawl samples (Table 2) in areas of high (Region 1) and low (Region 2) egg density. We feel that sampling sardines from both regions leads to a better estimate of spawning biomass for the whole population in the large oceanic area from San Diego to San Francisco. We found that, although average mature female weight (W) was similar in both regions (67 and 64 grams, Table 3), the spawning fraction, *S*, (based on females that spawned the night before capture, night *B* or "Day 1") was much higher in Region 1 (0.213 females per night) than in Region 2 (0.033 females per night). We also noticed a high fraction of immature sardine females (45% of 149 females) in Region 2 whereas only 9% of 80 females in Region 1 were immature. We recommend that future biomass surveys sample sardines in both high and low egg density areas.

We estimated that 50% of the female sardines were mature (ML_{50}) at 152 mm during April 2005 (Figure 8). The April 2005 estimate of ML_{50} is much lower than the 2004 value (193 mm) and about the same as the estimate from 1994 (159 mm) (Lo et al. 2005). The variation in ML_{50} could be real due to change in maturity or it may be the result of sample bias from one or more of the following: a) sardines were from the high egg density area only, b) all or a majority of the sardines were from offshore, c) all or a majority of the sardines were from inshore or near islands, d) migration of sardine subpopulations, and e) age and length relationship. We recommend continued evaluation of maturity to eliminate any biases.

Trawling during the 2005 survey covered a broad area and for the first time we were able to examine female parameters in offshore and inshore areas during the same time period. Ten trawls were considered "offshore", of which six were positive for sardines; nine trawls were conducted "inshore" or near islands and eight were positive (Table 6). We found that female sardines were larger offshore and a lower proportion of these randomly selected females were immature. In addition, mature females offshore were spawning at a higher fraction and fewer were classed as postbreeding (Macewicz et. al. 1996) than inshore (Table 6). These initial results not only suggest that larger females are offshore, but also that large females may be active for longer duration and may spawn more frequently. We recommend that future biomass surveys sample sardines both offshore and inshore.

Spawning biomass

The 2005 estimate of spawning biomass is considerably higher than that in 2004 and close to that in 2001. These differences are primarily due to the change of both the egg production and the adult reproductive output. The egg production, 1.916 eggs/0.05m^2, was higher than 0.96 eggs/0.05m^2 in 2004, 1.52 eggs/0.05m^2 in 2003 and 0.728 eggs/0.05m^2 in 2002, while the area of Region 1 of 46,203 km^2 was smaller than 68,204 km^2 in 2004, 83,578 km^2 in 2003 and 88,403 km^2 in 2002 (Table 4). The change in adult reproductive output to a lower daily specific fecundity (15.64 eggs/g/day) in 2005 than 2004 (21.86 eggs/g/day) was the result of the shift to smaller average females in the population and taking trawl samples from both Region 1 (the high density area) and Region 2 (the low density area). If the daily specific fecundity in Region 2 is substantially lower than that in Region 1 and the estimates of adult reproductive outputs are based on data from Region 1 only (Table 3), the spawning biomass will be underestimated. This may be true for 2002 and 2004 because only Region 1 was trawled. Therefore DEPM spawning biomass may have been underestimated in previous years (Lo and Macewicz 2004). The underestimation was more evident when the DEPM estimates were compared to spawning stock biomass (SSB) estimates based on the 2005 stock assessment (Hill et al. 2005, Lo et al. 2005) as most of the SSB in 1994-2005 were higher than the estimates of DEPM spawning biomass except for years 2000, 2001 and 2005 (Figure 9).

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Table 1. Number of positive tows of sardine eggs from CalVET, yolk-sac larvae from CalVET and Bongo, and eggs from CUFES in Region 1 (eggs/min ∃1) and Region 2 (eggs/min <1) for both *New Horizon* (NH) and *Jordan* (Jord) cruises 0504.

		Region 1			Region 2			Total	NH	Jord
		Total	**NH**	**Jord**	**Total**	**NH**	**Jord**			
CalVET eggs	positive	18	10	8	5	3	2	23	13	10
	Total	26	15	11	48	28	20	74	43	31
CalVET yolk-sac	positive	10	7	3	6	6	0	16	13	3
	Total	23	14	9	51	29	22	74	43	31
Bongo yolk-sac	positive	11	11	0	22	22	0	33	33	0
	Total	15	15	0	86	60	26	101	75	26
CUFES eggs	positive	155	92	63	142	27	115	297	119	178
	Total	188	118	70	773	281	492	961	399	562

Table 2. Individual trawl information, sex ratio[a], and parameters for mature female *Sardinops sagax*, used in estimation of March-April 2005 spawning biomass.

COLLECTION INFORMATION									MATURE FEMALES						
				Location									Number spawning		
Region	Trawl No.	Month-Day	Time	Latitude °N	Longitude °W	Surface Temp. °C	Number of fish sampled	Proportion of females	Number analyzed	Body weight (g) Ave.	Weight without ovary (g) Ave.	Batch Fecundity Ave.	Night of capture	Night before capture	Number Females Adjusted[b]
1	12	4-06	19:49	34.902	121.957	14.5	2	0.526	1	210.00	192.87	66489	0	0	1
1	19	4-24	22:01	34.619	121.706	14.3	8	0.724	4	162.00	152.03	51119	0	1	5
1	18	4-24	19:21	34.542	121.879	14.3	50	0.624	23	85.54	82.21	24848	3	5	25
1	8	4-05	04:14	34.320	121.593	14.2	6	0.472	2	197.00	189.22	65115	2	0	0
1	2	4-01	04:24	33.035	117.843	15.6	50	0.465	25	42.50	40.77	9257	1	5	29
1	1	3-31	23:43	32.848	117.604	15.4	50	0.467	25	43.18	41.52	9537	0	9	34
2	16	4-22	23:07	37.257	124.313	13.2	29	0.440	11	94.23	92.26	28632	0	2	13
2	14	4-11	20:36	36.919	122.540	12.2	50	0.516	16	74.03	73.16	21443	0	0	16
2	9	4-05	18:03	34.671	121.011	14.8	50	0.428	6	44.42	43.97	10461	0	0	6
2	10	4-05	21:21	34.529	121.070	14.5	50	0.352	19	55.92	54.18	14303	0	0	19
2	11	4-06	02:26	34.505	121.221	14.4	50	0.412	22	54.84	53.27	13961	0	0	22
2	6	4-03	22:41	34.215	120.231	12.9	15	0.738	7	42.50	41.70	9605	0	0	7
2	7	4-04	03:04	34.004	120.610	13.0	50	0.497	2	30.75	30.30	5315	0	0	2
2	3	4-01	19:28	33.025	119.484	15.2	47	0.292	12	79.79	76.11	22555	6	1	7

[a]Sex ratio, proportion of females by weight, based on average weights (Picquelle and Stauffer 1985).
[b] Required to estimate the fraction of mature females that spawned the night before capture if the number of females spawning on the night of capture is an overestimate (equation 9 of Picquelle and Stauffer 1985).

Table 3: Egg production (P_0) of Pacific sardine in 2005 based on egg data from CalVET and yolk-sac larval data from CalVET and bongo in Region 1 (eggs/min ≥1) and Region 2 (eggs/min <1) from *New Horizon* (Apr. 15-May 1) and *Jordan* (Mar. 28-Apr. 26) cruises, adult parameters from the *Jordan* cruise, and 2005 spawning biomass estimate. For comparison, spawning biomass estimates are given using 2004, 2002, and 1994 adult parameter data.

Parameter	**Region 1**	**Region 2**	**Whole area**			
number of pump samples (CUFES)	188	773	**961**	961	961	961
n: CalVET	26	48	**74**	74	74	74
P_0 / 0.05m^2 CalVET	8.14[a]	0.53	**1.92**	1.92	1.92	1.92
CV	0.4	0.69	**0.42**	0.42	0.42	0.42
Area (km^2)	46203	207417	**253620**	253620	253620	253620
%	18.22	81.78	**100**	100	100	100
Year for adult samples	2005	2005	**2005**	2004	2002	1994
Female fish wt (W)	67.02	63.93	**65.34**	166.99	159.25	82.5
Batch fecundity (F)	18032	17440	**17662**	55771	54403	24283
Spawning fraction (S)	0.213	0.033	**0.1236**	0.131	0.1739	0.149[b]
Sex ratio (R)	0.550	0.425	**0.468**	0.5[c]	0.386	0.537
Eggs/g biomass/day (RSF/W_f)	31.498	3.76	**15.666**	21.852	22.9315	23.55
Spawning biomass (mt)[d]	238800	584306	**621508**	445680	424700	413546
CV			**0.54**			
Daily mortality (Z)	0.579					
CV	0.2					
eggs/min	3.1	0.067	**0.62**			
CV	0.16	0.57	**0.15**			
q = eggs/min in Region 2/eggs/min in Region 1			**0.065**			
CV			**0.61**			
E = eggs.min/eggs/tow			**0.18**			
CV			**0.28**			
n: bongo	15	86	**101**			
Area in nm^2	13499.8	60603.9	**74103.7**			
S. biomass (ston)			**683657**	490248	467170	454901

P_0/0.05m^2 was from CalVET only for 2001 and beyond

[a] 8.14 was corrected for bias of P_0.

[b] average spawning fraction of active females collected 1986-1994 (Table 8 Macewicz et al. 1996)

[c] used because authors were uncertain if biases were affecting 2004 calculated ratio of 0.618

[d] biomass was computed from estimates of parameters in each column, i.e. 2005 whole area is an average of adult parameters and 621508 ≠ 238800 + 584306

The estimate of spawning biomass in this table may be different from Table 5 due to rounding error.

Table 4. Estimates of daily egg production (P_0) for the survey area, daily instantaneous mortality rates (Z) from high density area (region 1), daily specific fecundity (RSF/W), spawning biomass of Pacific sardine and average sea surface temperature for the years 1994 to 2005.

Year	P_0 (CV)	Z (CV)	Area (km^2) (Region 1)	$\frac{RSF}{W}$	Spawning biomass (mt)[b](CV)	Ave. Temp. for positive egg or yolk-sac samples (°C)	Mean temperature (°C)	Methods for P_0
1994	0.193 (0.21)	0.120 (0.91)	380,175 (174,880)	11.38	127,102 (0.32)	14.3	14.7	Weighted nonlinear on grouped data
1995	0.830 (0.5)	0.400 (0.4)	113,188.9 (113,188.9)	23.55[a]	79,997 (0.6)	15.5	14.7	Weighted nonlinear on original data
1996	0.415 (0.42)	0.105 (4.15)	235,960 (112,322)	23.55	83,176 (0.48)	14.5	15.0	Composite estimate
1997	2.770 (0.21)	0.350 (0.14)	174,096 (66,841)	23.55	409,579 (0.31)	13.7	13.9	Weighted nonlinear on grouped data
1998	2.279 (0.34)	0.255 (0.37)	162,253 (162,253)	23.55	313,986 (0.41)	14.38	14.6	Composite estimate
1999	1.092 (0.35)	0.100 (0.6)	304,191 (130,890)	23.55	282,248 (0.42)	12.5	12.6	Composite estimate
2000	4.235 (0.4)	0.420 (0.73)	295,759 (57,525)	23.55	1,063,837 (0.67)	14.1	14.4	Composite estimate
2001	2.898 (0.39)	0.370 (0.21)	321,386 (70,148)	23.55	790,925 (0.45)	13.3	13.2	Weighted nonlinear on original data
2002	0.728 (0.17)	0.400 (0.15)	325,082 (88,403)	22.94	206,333 (0.35)	13.6	13.6	Weighted nonlinear on original data
2003	1.520 (0.18)	0.480 (0.08)	365,906 (82,578)	22.94	485,121 (0.36)	13.7	13.8	Weighted nonlinear on original data
2004	0.960 (.24)	0.250 (0.04)	320,620 (68,234)	21.86	281,639 (0.3)	13.4	13.7	Weighted nonlinear on original data
2005	1.916 (0.417)	0.579 (0.20)	253,620 (46,203)	15.67	621,657 (0.54)	14.21	14.1	Weighted nonlinear on original data

[a] from Table 3: computation for 1994 after setting S to 0.149 (the average spawning fraction of active females from 1986-1994; Macewicz et al. 1996)

[b] $CV(B_s)=(CV^2(P_0)+allotherCOV^2)^{1/2}=(CV^2(P_0)+0.054)^{1/2}$. For years 1995-2001 allotherCOV^2 was from 1994 data.(Lo et al. 1996). For year 2003, allotherCOV was from 2002 data (Lo and Macewicz 2002)

Table 5. The 2005 output from "frmBIOMASS" form in the EPM program after input of 2005 parameters and estimation of adult parameters (top box)**,** and 'Estimate Correlation and Biomass also' (bottom box) (Appendix II Chen et al. 2003).

EPM PROGRAM

Calculate Correlation, Biomass and CV

Input Parameters:

Enter first collection number	14243
Enter last collection number	14261
Enter first collection number of 2nd set	
Enter last collection of 2nd set	
Mean gonad-free weight used in regression	89.7
Number of fish used in regression	22
Variance about regression	23234053
Variance of regression slope	390.46
Po (eggs/day - 0.05m2)	1.92
Variance of Po	0.638
Area (square kilometers)	253620

Statistic Results:

	Average	Variance
Whole Body Weight	65.338997377	48.186347336
Gonad Fee Weight	63.108964	44.718614881
Batch fecundity	17661.640974	8703856.9576
Spawners, Day 0	0.0685714286	0.0014766016
Spawners Day 1	0.123655914	0.0014683322
Sex Ratio	0.4686950598	0.0011762344
Daily specific fecundity	15.666206759	
Number of Sets	14	

Enter the name of DBSET table	DBSET05
Estimate Average and Variance only	Estimate
Estimate Correlation and Biomass also	Estimate

EGG PRODUCTION VALUES

Po (eggs/day - .05 m2):	1.92
CV of Po:	0.41601511558
Area (square kilometers):	253620

BIOMASS ESTIMATE

Biomass (m - tons) =	621657.057775
CV (biomass) =	0.54040739276

CORRELATIONS

Parameter	W	F	S	R
Whole - Body Weight (W)		0.85233114	-0.1589059	0.30195276
Batch Fecundity (F)			-0.1512194	0.25719435
Fraction Spawning (S)				0.27014866
Sex Ratio (R)				

Table 6. Pacific sardine female parameters by location during March-April 2005 survey.

Parameters	Location: Inshore	Location: Offshore
Trawl number	1, 2, 6, 7, 9, 10, 11, 14	3, 8, 12, 16, 18, 19
Number of females	172	57
Standard length (mm)		
Mean	158	197
Range	121 - 195	161 - 265
Whole body weight (g)		
Mean	44.6	97.4
Range	18.5 - 95.5	50.0 - 210.0
Proportion Immature (%)	40.7	7.0
Average fraction of mature [a] spawning	0.064	0.189
spawning the night of capture	0.010	0.208
spawned the night before capture	0.118	0.170
Average fraction of active [b] spawning	0.080	0.209
spawning the night of capture	0.012	0.229
spawned the night before capture	0.148	0.188
Proportion of postbreeding [c] mature (%)	20.6	9.4

[a] number of mature females includes all active and post-breeding females.

[b] active females are capable of spawning and have ovaries containing oocytes with yolk or postovulatory follicles < 60 hours old.

[c] postbreeding females are mature females that are no longer active and considered incapable of further spawning in the season (Macewicz et el. 1996).

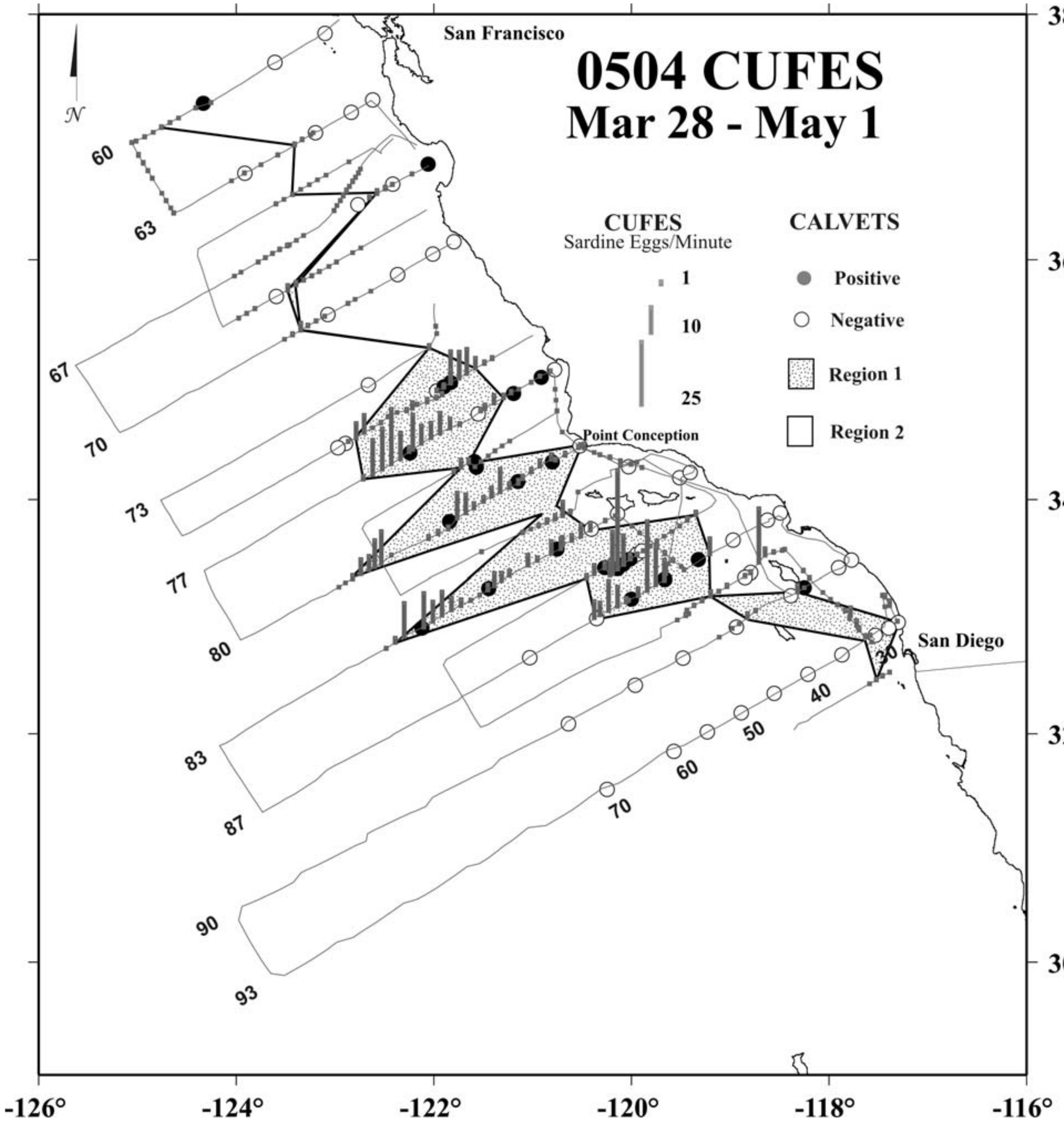

Figure 1. Sardine eggs from CalVET, a.k.a. Pairovet; (solid circle denotes positive catch and open circle denotes zero catch) and from CUFES (stick denotes positive collection) in March-April 2005 survey. The numbers on line 93 are CalCOFI station numbers. Region 1 is stippled area.

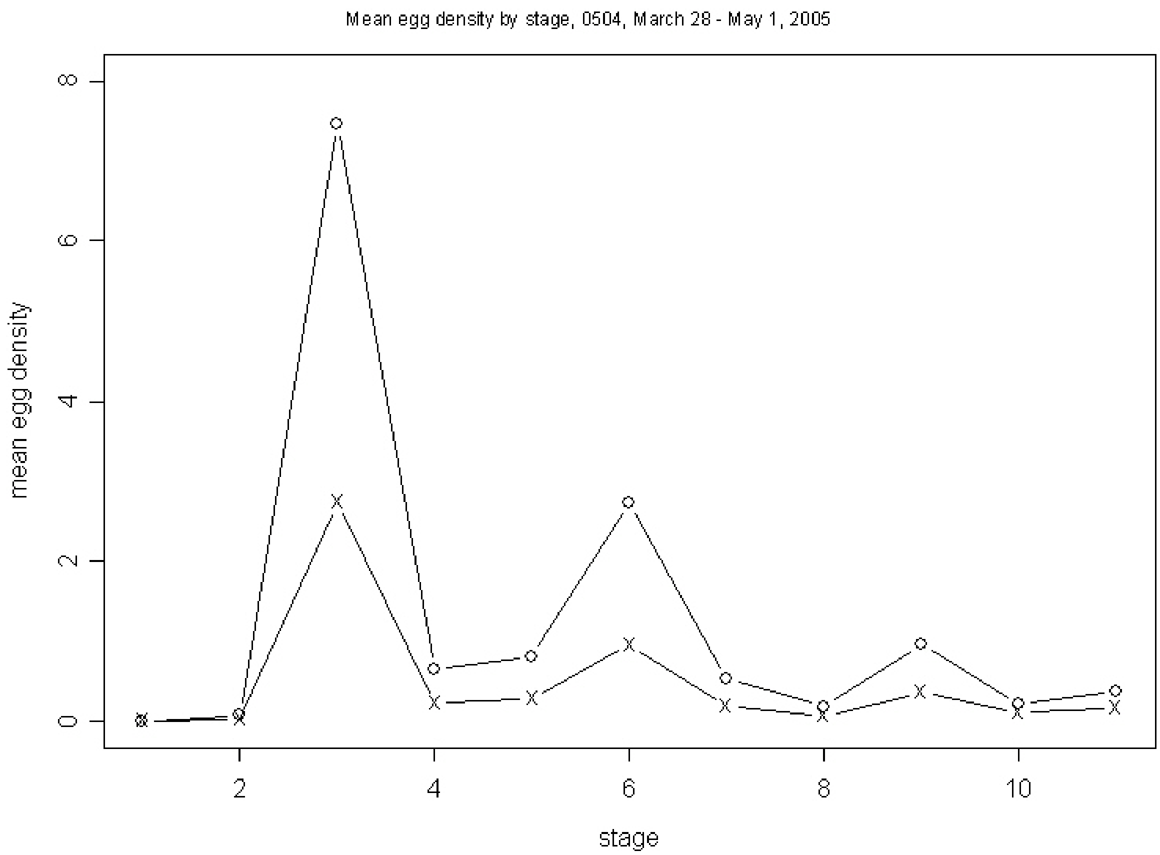

Figure 2. Sardine eggs per 0.05 m^2 for each developmental stage for March - April, 2005. Symbols: o = Region 1 and x = entire survey area.

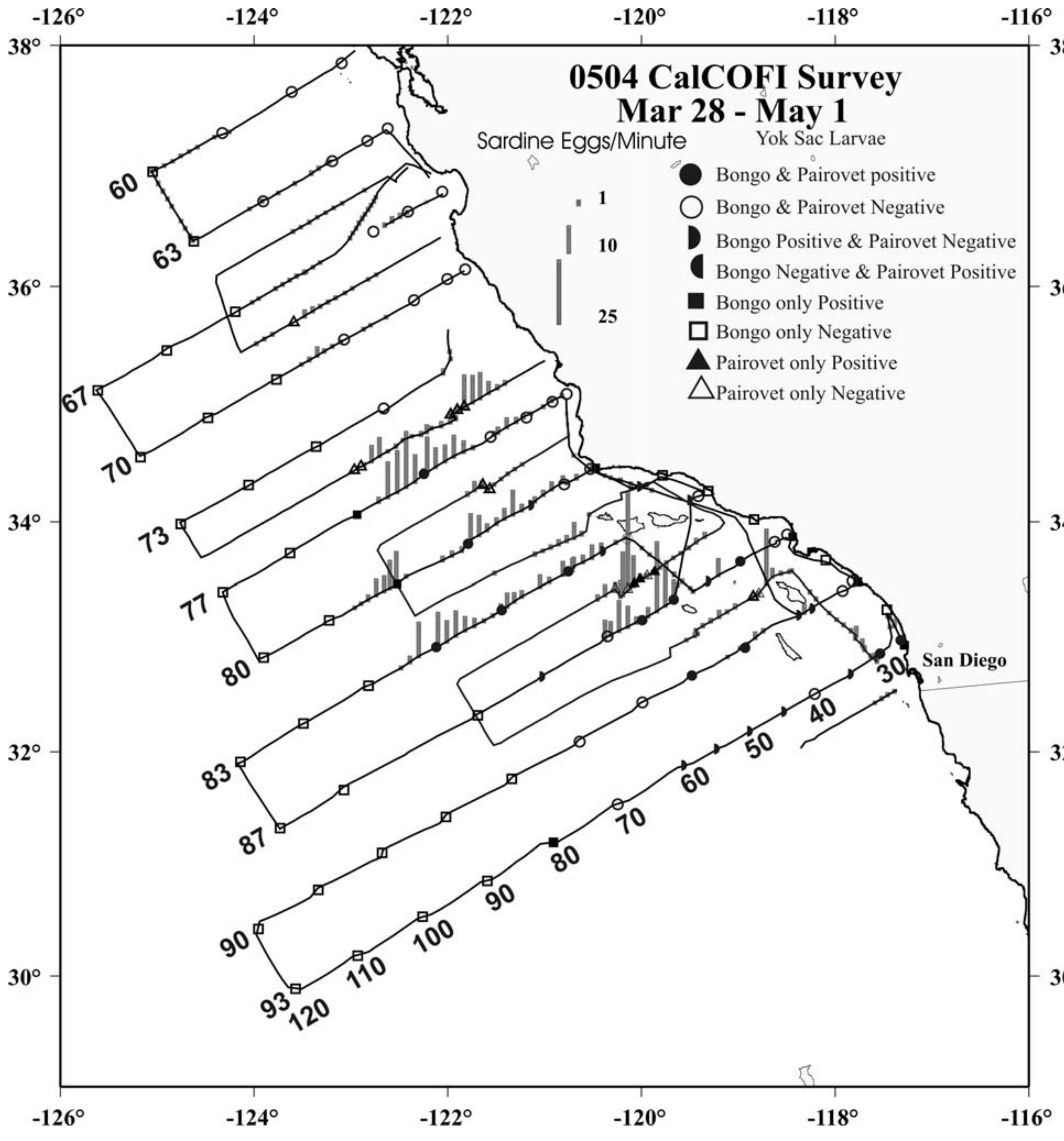

Figure 3. Sardine yolk-sac larvae from CalVET (or Pairovet; circle and triangle) and from Bongo (circle and square) in March-April 2005 survey. Solid symbols are positive and open symbols are zero catch.

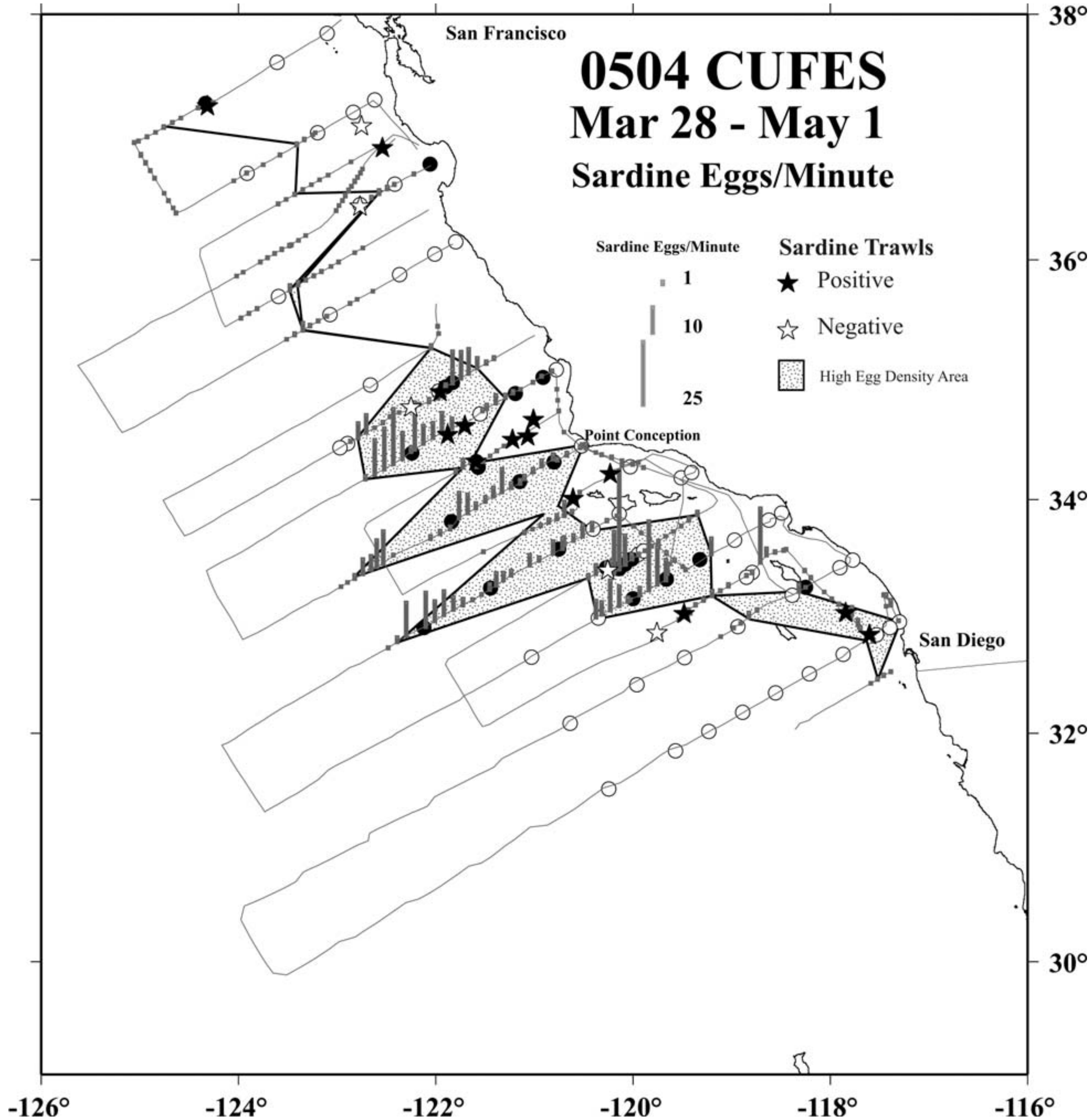

Figure 4. March-April 2005 trawl start locations. Solid stars are trawls that contained at least one sardine. Background is CUFES track lines and sardine egg abundance, pairovet tows (circles: solid are positive for sardine eggs or larvae; open are negative). The high egg density area is Region 1.

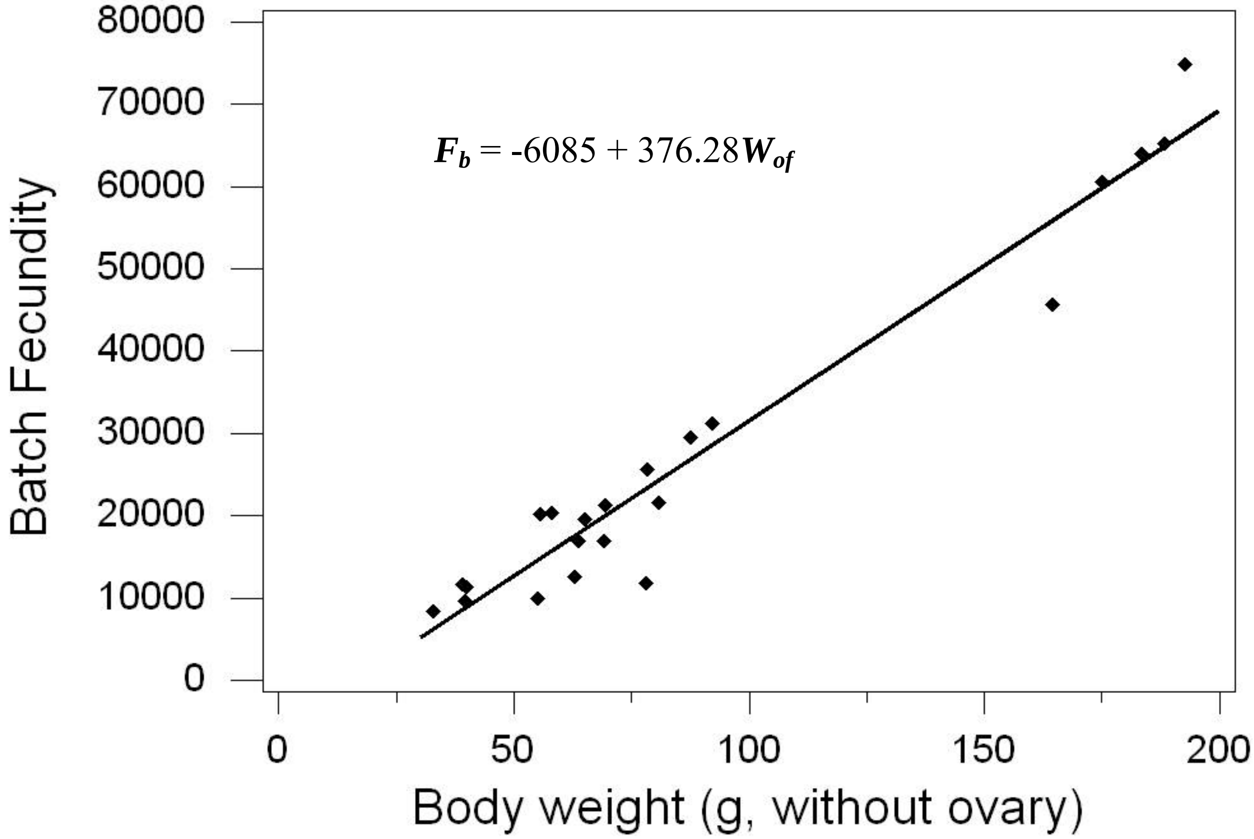

Figure 5. Batch fecundity (F_b) of *Sardinops sagax* as a function of female body weight (W_{of}, without the ovary) for 22 females taken during April 2005. The batch was estimated from numbers of hydrated or migratory-nucleus-stage oocytes.

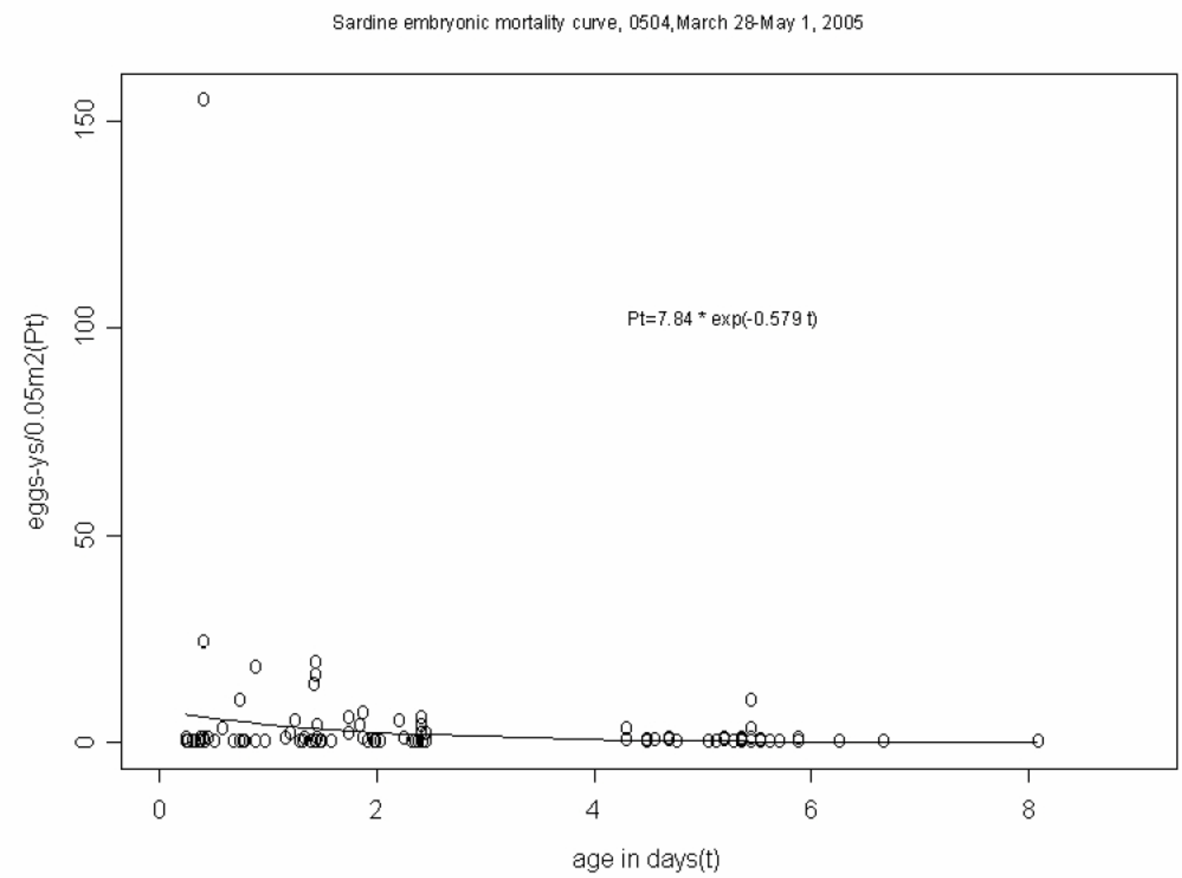

Figure 6. Embryonic mortality curve of Pacific sardine. Staged egg data were from CalVET and yolk-sac larval data were from CalVET and Bongo in 2005. The number, 7.84, is the estimate of daily egg production before correction for bias.

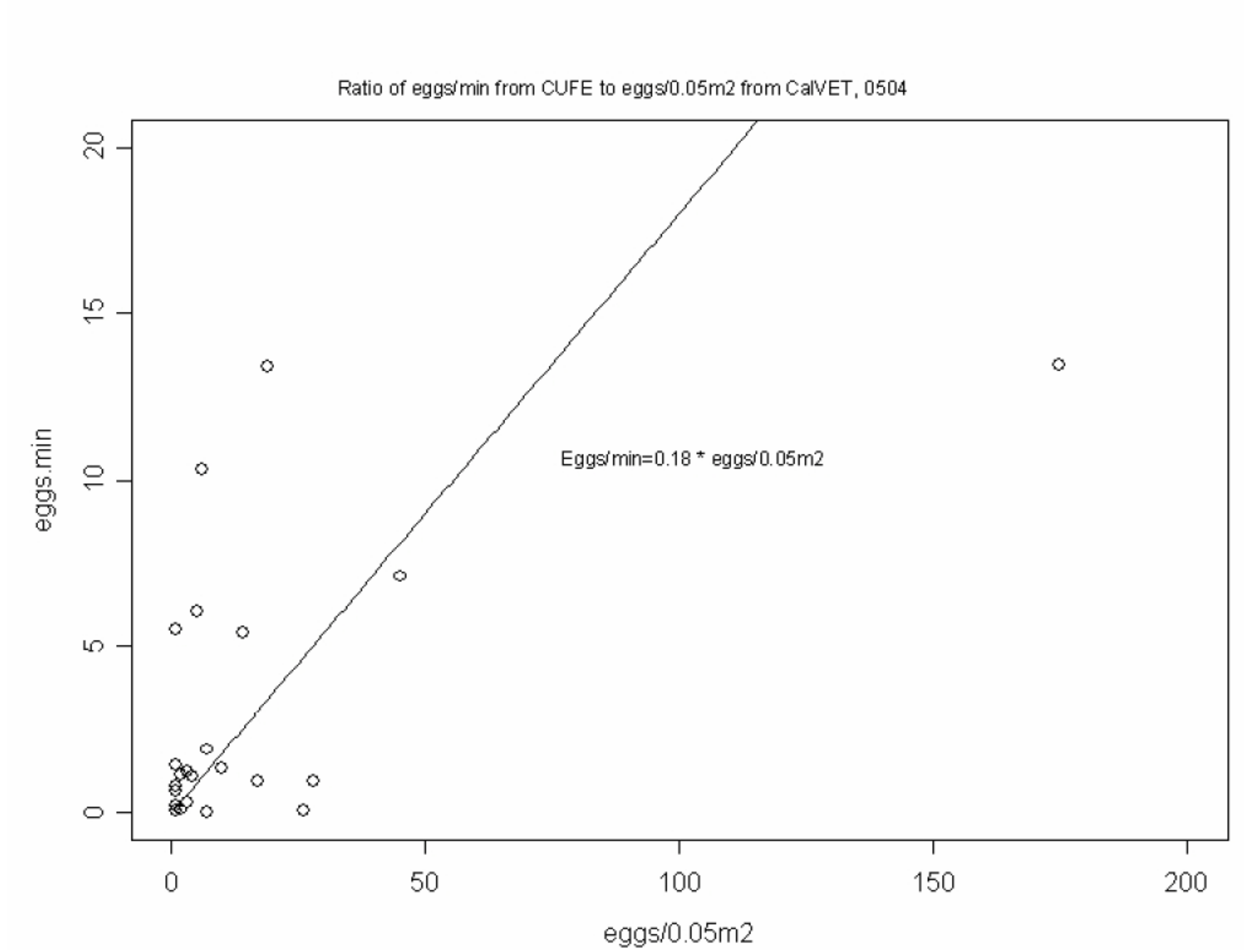

Figure 7. Catch ratio of eggs/min from CUFES to eggs/0.05m^2 from CalVET during March-April 2005 excluding the data point of 178 eggs.

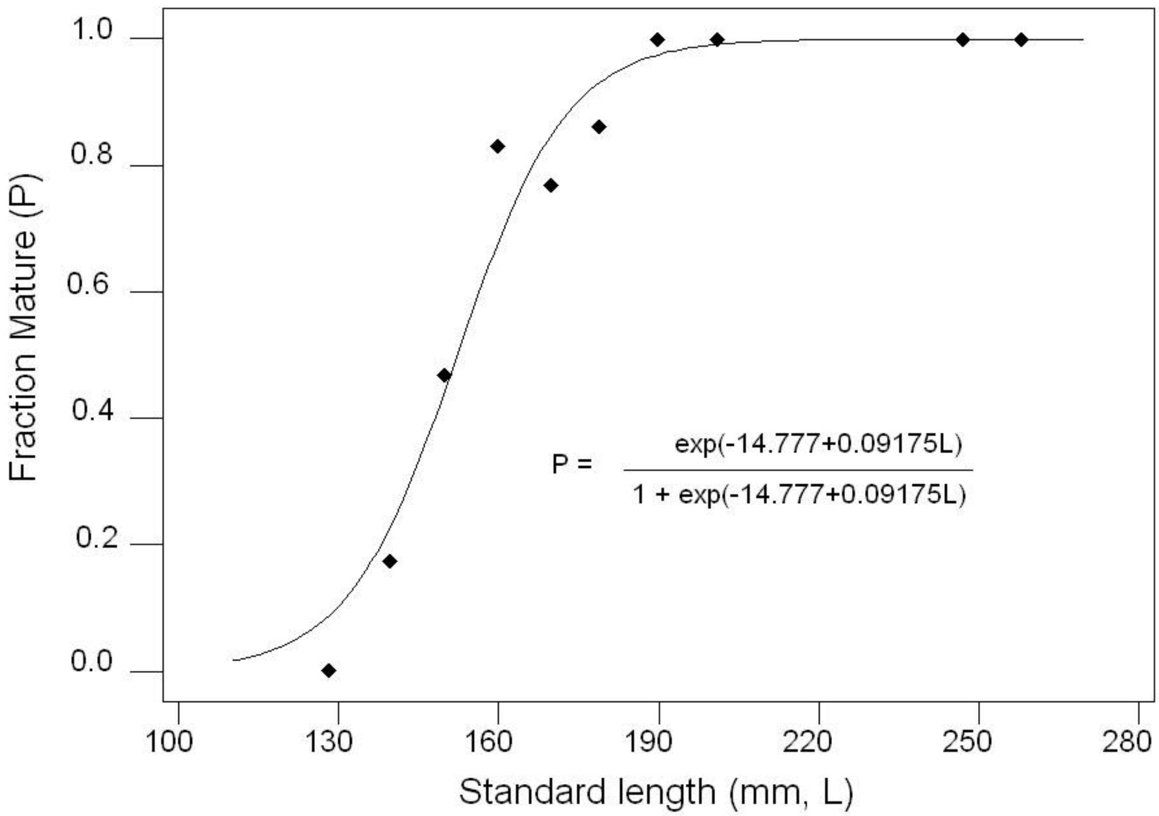

Figure 8. Fraction of Pacific sardine females randomly sampled during April 2005 that were sexually mature as a function of standard length. Symbols represent actual fraction mature within 10 mm length classes.

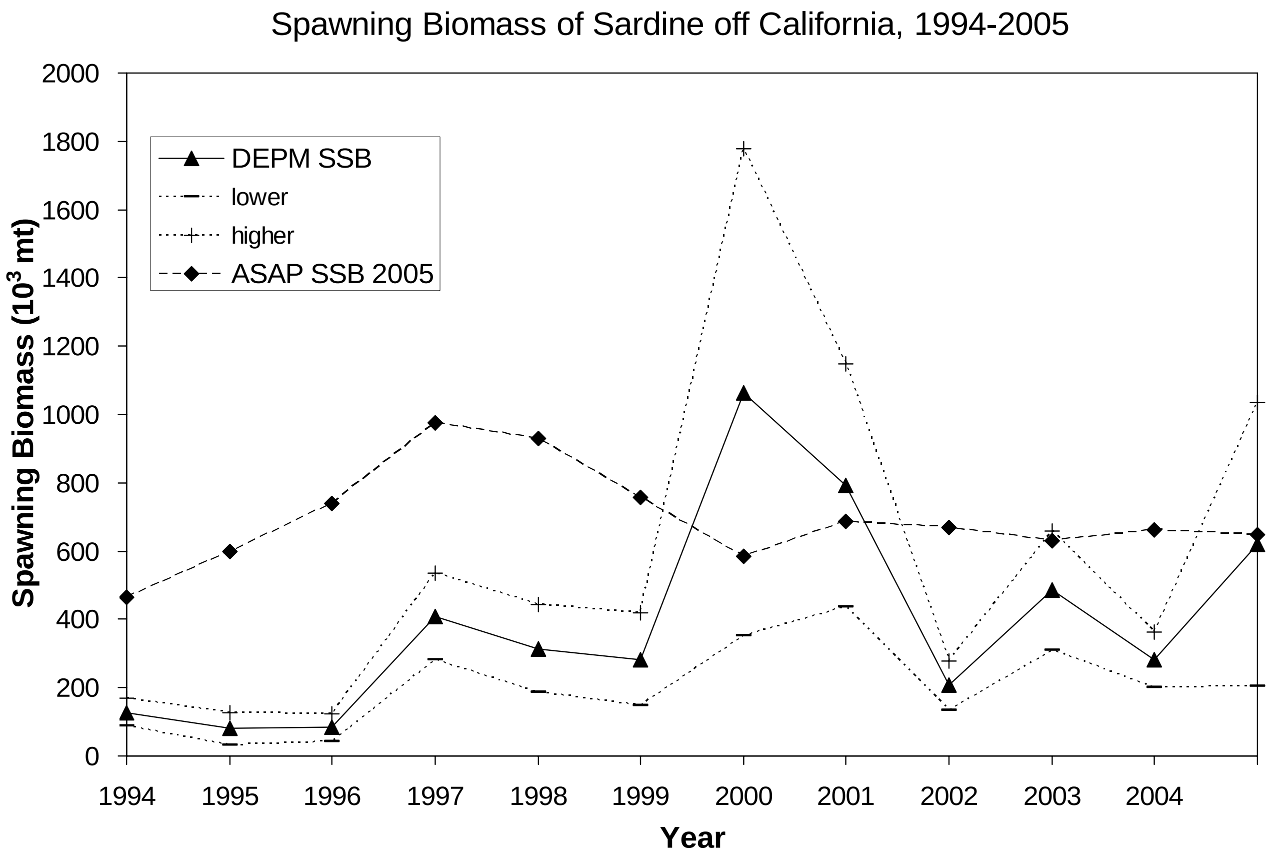

Figure 9. Spawning biomass of Pacific sardine off California from 1994-2005, estimated from the DEPM with lower limit (DEPM estimates -1SE) and higher limit (DEPM estimates +1SE), compared to the estimates of the spawning stock biomass (SSB) from the ASAP (Age structured assessment program) based on data from 1985-2005 (ASAP SSB 2005).

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